

CLAIM AMENDMENTS

Please amend the claims as follows:

1-12. (Cancelled)

13. (Currently Amended) A method of production of stratified, terminally-differentiated human ~~mammalian~~ urothelium in which urothelial cells, isolated from the ~~mammalian~~ human body and propagated by culture in serum-free medium, are transferred to passaged through a first nutrient differentiation medium containing serum and then redispersed by passage before being added to a second nutrient differentiation medium containing serum to form said urothelium, wherein the first nutrient differentiation medium is not the second nutrient differentiation medium.

14. (Cancelled)

15. (Previously Presented) The method of claim 13 in which the serum is bovine serum.

16. (Currently Amended) The method of claim 15 in which the serum is adult or fetal bovine serum.

17. (Currently Amended) The method of claim 13 in which the concentration of ~~the components of the~~ serum as a proportion of the final volume of the first or second nutrient differentiation medium is between about 1% and about 30% related to the concentration of ~~said~~ components in whole serum.

18. (Currently Amended) The method of claim 13 in which the concentration of ~~the components of the~~ serum as a proportion of the final volume of the first or second nutrient differentiation medium is between about 3% and about 10% related to the concentration of ~~said~~ components in whole serum.

19. (Currently Amended) The method of claim 13 wherein the concentration of the ~~components of~~ the serum as a proportion of the final volume of the first or second nutrient differentiation medium is between about 4% and about 6% related to the concentration of said components in whole serum.

20. (Currently Amended) The method of claim 13 wherein the first or second nutrient differentiation medium is, or is a derivative of, MCDB-153 medium.

21. (Currently Amended) The method of claim 13 wherein the first or second nutrient differentiation medium is KSFM (Keratinocyte Serum Free Medium).

22. (Currently Amended) The method of claim 13 wherein the first or second nutrient differentiation medium is supplemented by one or more of Epidermal Growth Factor (EGF); Bovine Pituitary Extract (BPE); or Cholera Toxin (CT).

23. (Previously Presented) Urothelium produced by the method of claim 13.

24. (Currently Amended) A method of production of stratified, differentiated ~~mammalian~~ human urothelium, the method comprising:

~~disaggregating cells of a primary serial~~ culture of ~~mammalian~~ human urothelial cells in a serum-free nutrient medium;

replacing the serum-free nutrient medium with ~~dispersing the urothelial cells of the primary culture into~~ a first differentiation cell culture medium that includes whole serum;

~~culturing~~ maintaining the urothelial cells in the first differentiation culture medium to form a ~~secondary~~ cell culture having aggregated urothelial cells;

dispersing and disaggregating the aggregated urothelial cells into a second differentiation cell culture medium that includes whole serum, wherein the first differentiation cell culture medium is not the second differentiation cell culture medium; and

culturing the urothelial cells in the second differentiation culture medium so as to form stratified, terminally-differentiated ~~mammalian~~ human urothelium.

25. (Previously Presented) A method as in claim 24, wherein the aggregated urothelial cells are at least partially confluent.

26. (Previously Presented) A method as in claim 24, wherein the aggregated urothelial cells approach confluency.

27. - 28. (Cancelled)

29. (Previously Presented) A method as in claim 24, wherein the serum is at a concentration between about 1% and about 30% of the medium.

30. (Previously Presented) A method as in claim 24, wherein the serum is at a concentration between about 4% and about 6% of the medium.

31. (Previously Presented) A method as in claim 24, wherein the first differentiation, and/or second differentiation cell culture medium is one of MCDB-153 medium, KSFM (Keratinocyte Serum Free Medium), or a medium derived thereof.

32. (Previously Presented) A method as in claim 24, wherein first differentiation, and/or second differentiation cell culture medium is supplemented by at least one of Epidermal Growth Factor (EGF), Bovine Pituitary Extract (BPE), or Cholera Toxin (CT).

33. (Previously Presented) A method as in claim 24, wherein the culturing includes increasing the calcium concentration in the second differentiation cell culture medium.

34. (Currently Amended) A method of production of stratified, differentiated ~~mammalian~~ human urothelium, the method comprising:

~~disaggregating cells of a primary culture of mammalian human urothelial cells in~~
~~a serum-free nutrient medium;~~

~~replacement of the serum-free nutrient medium with dispersing the urothelial cells~~
~~of the primary culture into a first differentiation low calcium cell culture medium that~~
includes at least 5% whole serum;

~~culturing maintaining~~ the urothelial cells in the first differentiation cell culture
medium to form a secondary cell culture having aggregated urothelial cells;

dispersing and disaggregating the aggregated urothelial cells into a second
differentiation ~~low calcium~~ cell culture medium that includes at least 5% whole serum;
and

culturing the urothelial cells and increasing the calcium concentration of the
second differentiation ~~third~~ culture medium so as to form stratified, terminally-
differentiated ~~mammalian human~~ urothelium.

35. (New) A method as in claim 34, further comprising determining the
urothelial cells from the third culture medium to have stratified layers of terminally-
differentiated human urothelium.